Kunio MITUI*: Spore ornamentations of Japanese species of *Lepisorus*

三井邦男*: 日本産ノキシノブ属の胞子表面の模様について

(Plates XVI—XIX)

The spores of six species of Japanese *Lepisorus* have been studied by Kawasaki microscopically. They are the spore of B_2 type (bilateral with small verrucae or rough exospore, surface yellow or orange or brown, length 28 μ–80 μ) in his system, and there are some differences in their ornamentations. From his description two types of spore ornamentation are found among them; one is the spore with rough exospore, and the other has muriferous exospore.

I have reported already some cytological and morphological characters of Japanese *Lepisorus* (except for the spore characters) and have concluded that this genus might be separated into two phylogenetic groups; one is based on basic chromosome number 25 or 26 and the other is X=35.

In the present study, I observed the spore ornamentations of nine species in Japanese *Lepisorus* by the scanning electron microscope in attempt to clarify that there are fundamental differences in spore ornamentation between the above mentioned phylogenetic groups.

I am greatly indebted to Assist. Prof. Tsugio Kawasaki, Tokyo Gakugei University for his helpful advice. For technical help with the scanning electron microscope, I thank Miss Yoshiko Tanaka of Nippon Dental College.

Material and method

The spores were collected from the following specimens as shown in Table 1.

The preparation of materials for the scanning electron microscope was made as the following procedures: Matured spores were put on the speci-

* Department of Biology, Nippon Dental College. Fujimi, Chiyoda-Ku, Tokyo, Japan. 日本歯科大学生物学教室

Table 1. Localities of specimens.

Species	Locality
L. angustus	Okukinu, Tochigi Pref.
L. annuifrons	Kawamata, Saitama Pref.
L. clathratus	Mt. Toyoguchi, Nagano Pref.
L. hachijoensis	Hachijo Isl., Tokyo Pref.
L. onoei	Mt. Hakkōda, Aomori Pref.
L. thunbergianus (2X)	Mugi, Kaifu-Gun, Tokushima Pref.
〃 (4X)	Iwayadera, Ehime Pref.
L. tosaensis	Mugi, Kaifu-Gun, Tokushima Pref.
L. uchiyamae	Mugi, Kaifu-Gun, Tokushima Pref.
L. ussuriensis var. *distans*	Mt. Tōgatake, Kanagawa Pref.

men stub covered with the appropriate mountant (Do-tite) and they were coated with only gold metal. The gold metal wire was evaporated in a vacuum of 10^{-5} Torr. The young spores were fixed in 1 : 3 acetic alcohol and dehydrated in the following mixture: 1 : 1 acetic alcohol, 1 : 3 acetic alcohol, 1 : 9 acetic alcohol, 99% alcohol and absolute alcohol. These were also coated with only gold metal. The scanning electron microscope used was the JSM-U3 using accelerator voltage 15KV.

Observations

1. The development of the spore ornamentation in *L. thunbergianus*

The development of the spore ornamentation was observed as the following processes in the diploid of *L. thunbergianus*. In the early stage (when the nucleus of the tetrad spores can be observed through the sporoderm by light microscope), the sporoderm was coverded densely with small granuloid ornamentations (Pl. XVI, A, B, C). The same kind of these ones might be observed also in the matured spores of *Pleurosoriopsis makinoi*, whose spores had the transparent exine (Pl. XVI, D). According to the enlarging of spores, some foveolate ornamentations (I mean some hollowed places) could be seen on the surface of the sporoderm, when the granules were not remarkable ones (Pl. XVI, E, F, G). In this stage, the nucleus of spore had not been observed yet through the sporoderm by light microscope. Gradually, these foveolate ornamentations became remarkable ones on the

whole surface, therefore, from the reverse meaning, the faint reticulate ornamentation could be observed (Pl. XVI, H) in this case. Subsequently, some pilate processes appeared in somewhere on the spore surface and gradually these processes developed and further they were combined together to construct the muri (Pl. XVII, A-E). And consequently the remarkable reticulum was able to see on the spore on which the muri developed on the whole surface of spore (Pl. XVII, F).

In the tetraploids, the developmental stage of spore ornamentation was similar to that of diploid, however few pilate processes were observed and the thickening of muri seemed to occur simultaneously on the spore surface. Furthermore, the reticulum of tetraploid species seemed to be closer than that of diploid (Pl. XVII, G, H).

2. The spore ornamentations of eight species of *Lepisorus*

The sculptures of eight species may be described as the followings.

a) *L. onoei* (Pl. XVIII, A, B) The spore ornamentation of this species was closely similar to that of young spore of *L. thunbergianus*. The development of muri was not so advanced and some pilate processes could be found on the spore surface.

b) *L. angustus* (Pl. XVIII, C, D). The muri of this spore developed more than that of *L. onoei* and their reticula became to be more remarkable.

c) *L. hachijoensis* (Pl. XVIII, E, F). The ornamentation was similar to that of *L. angustus*, however more pilate processes were observed on the muri than *L. angustus*. Hence, this spore showed the rough surface and the reticulum of this spore was not so clear.

d) *L. ussuriensis* var. *distans* (Pl. XVIII, G, H). Any pilate processes were not observed on the surface of spore and the height of muri seemed to be equal in everywhere on the spore surface, therefore the surface of spore showed the smooth feature as described by Kawasaki. The reticulum of this spore was the clearest in eight species.

e) *L. annuifrons* (Pl. XVIII, A, B). The surface of this spore showed the reticulum feature and was similar to that of *L. ussuriensis* var. *distans*. However, the muri of this spore rised here and there on the spore surface and then the spore showed the roughly waved feature.

f) *L. uchiyamae* (Pl. XIX, C, D). The muri of this spore developed more than that of *L. annuifrons* and consequently the luminae of the reti-

culum became smaller than those of above mentioned species. Hence, the surface of this spore showed the rough feature as described by Kawasaki.

g) *L. tosaensis* (Pl. XIX, E, F). The muri of this species developed most closely on the whole surface of the spore and their rough characters were the strongest in nine species. The luminae of the reticulum was not so remarkable in this species.

h) *L. clathratus* (Pl. XIX, G, H). The muri developed uniformly on the whole surface of spores, but the height of muri was very low and was equal height on the spore surface as shown in *L. ussuriensis* var. *distans*. The reticulum was remarkable and the luminae became smaller than those of *L. uchiyamae*.

Discussion

As mentioned above, Kawasaki has observed the spore with rough exospore in *L. thunbergianus, L. onoei, L. tosaensis, L. clathratus* and *L. uchiyamae*. Also he has reported the spore with muriferous exospore in *L. ussuriensis* var. *distans* and *L. annuifrons*. However, in the present paper, it seems that each species of this genus has the spores with reticulum sculptures. Furthermore, it may be said that the differences among the species of this genus should be caused by the different development of the muri which constitute the reticulum on the spore surface. For example, the spores of *L. ussuriensis* var. *distans*, which are mentioned as having the smooth exospore by Kawasaki, have the smooth and continuous muri all over the spore surface and large remarkable luminae. On the other hand, the spores which have rough exospore in his description, have the well developed and discontinuous muri on the surface and small luminae of the reticulum.

The degree of the development of the muri seems to be related to the polyploidy in each species. For instance, *L. onoei* (n=25) had the spores whose muri were scarecely developed and only found several pilate processes on the spore surface. On the contrary, the most developed muri were observed in the spores of the species having the higher incidence of polyploidy such as *L. tosaensis* (n=75) and *L. uchiyamae* (n=70).

As far as the present study, the fundamental differences were not observed in the spore sculpture between two phylogenetic groups suggested in my previous paper.

References

Erdtman, O. G. E. 1957: Pollen and spore morphology and plant taxonomy. Stockholm. Kawasaki, T. 1964: Bulletin of Tokyo Gakugei University, 16: 69–74. Mitui, K. 1971: Journ. Jap. Bot. 46: 19–32.

Explanation of Plates XVI–XIX

Plate XVI. A. Tetrad of *L. thunbergianus*: N. Nucleus (×1820). B. Tetrads and young spore of *L. thunbergianus* by microscope: N. Nucleus (×400). C. Surface of tetrad of *L. thunbergianus* (×6500). D. Spore surface of *Pleurosoriopsis makinoi* (×1950). E. Young spore of *L. thunbergianus* (×650) (30°tilt). F, G and H. Surface of young spore of *L. thunbergianus* (×1950).

Plate XVII. The developmental process of spore ornamentation in *L. thunbergianus*. A.–F. Spores of diploid species of *L. thunbergianus* (×730). G and H. Spores of tetraploid species of *L. thunbergianus* (×730).

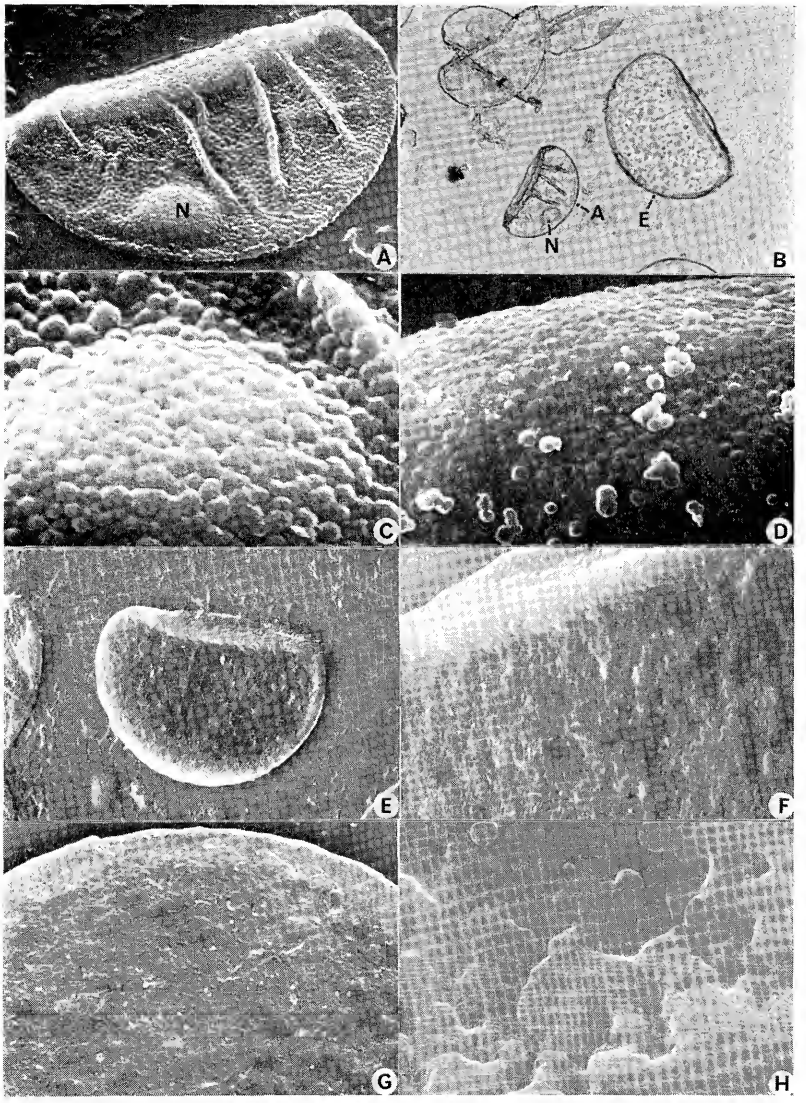
Plate XVIII. A. Spore of *L. onoei* (×650). B. Spore surface of *L. onoei* (×1950). C. Spore of *L. angustus* (×650). D. Spore surface of *L. angustus* (×1950). E. Spore of *L. hachijoensis* (×650). F. Spore surface of *L. hachijoensis* (×1950). G. Spore of *L. ussuriensis* var. *distans* (×650). H. Spore surface of *L. ussuriensis* var. *distans* (×1950).

Plate XIX. A. Spore of *L. annuifrons* (×650). B. Spore surface of *L. annuifrons* (×1950). C. Spore of *L. uchiyamae* (×650). D. Spore surface of *L. uchiyamae* (×1950). E. Spore of *L. tosaensis* (×650). F. Spore surface of *L. tosaensis* (×1950). G. Spore of *L. clathratus* (×650). H. Spore surface of *L. clathratus* (×1950).

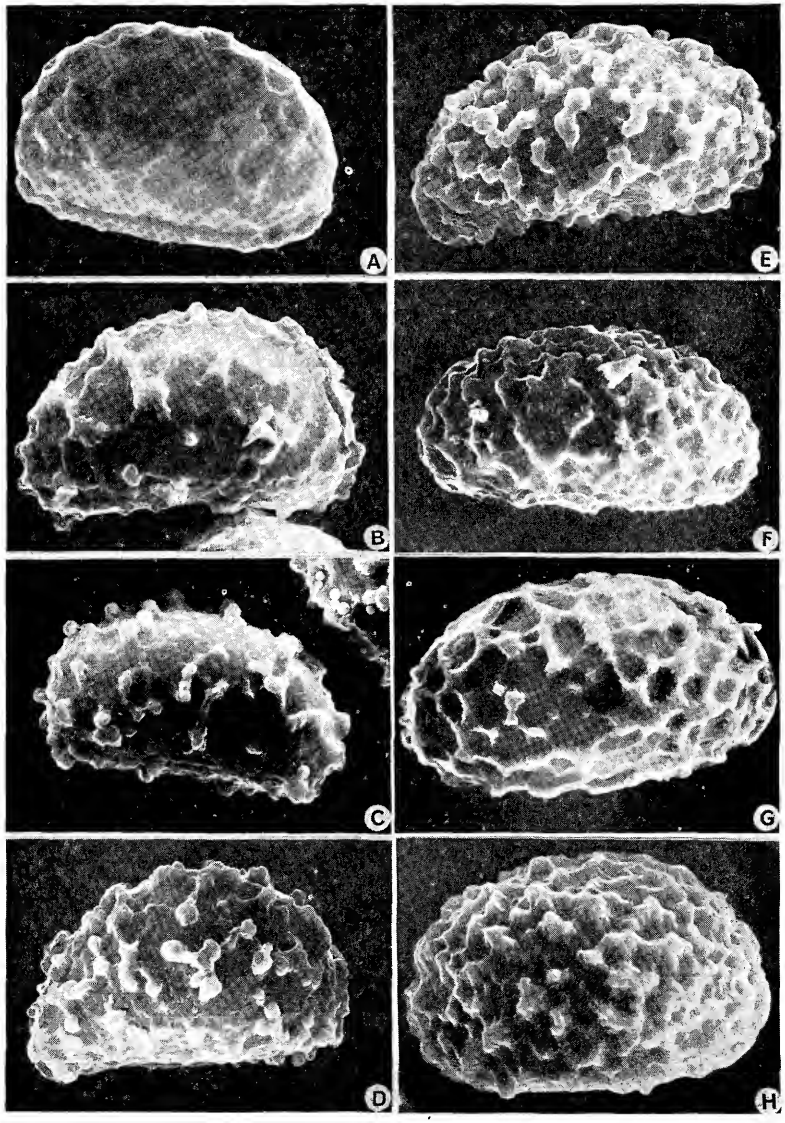
* * * *

日本産ノキシノブ属9種の胞子表面の模様を走査電子顕微鏡で観察した。9種とも表面の模様は網目状をしているが，ミヤマノキシノブ，ナガオノキシノブでは網目が顕著であるのに対し，ヒメノキシノブではうねがあまり発達しないので顕著ではない。またツクシノキシノブやコウラボシではうねが非常に発達し凹凸が大きくなり網目模様は顕著ではない。

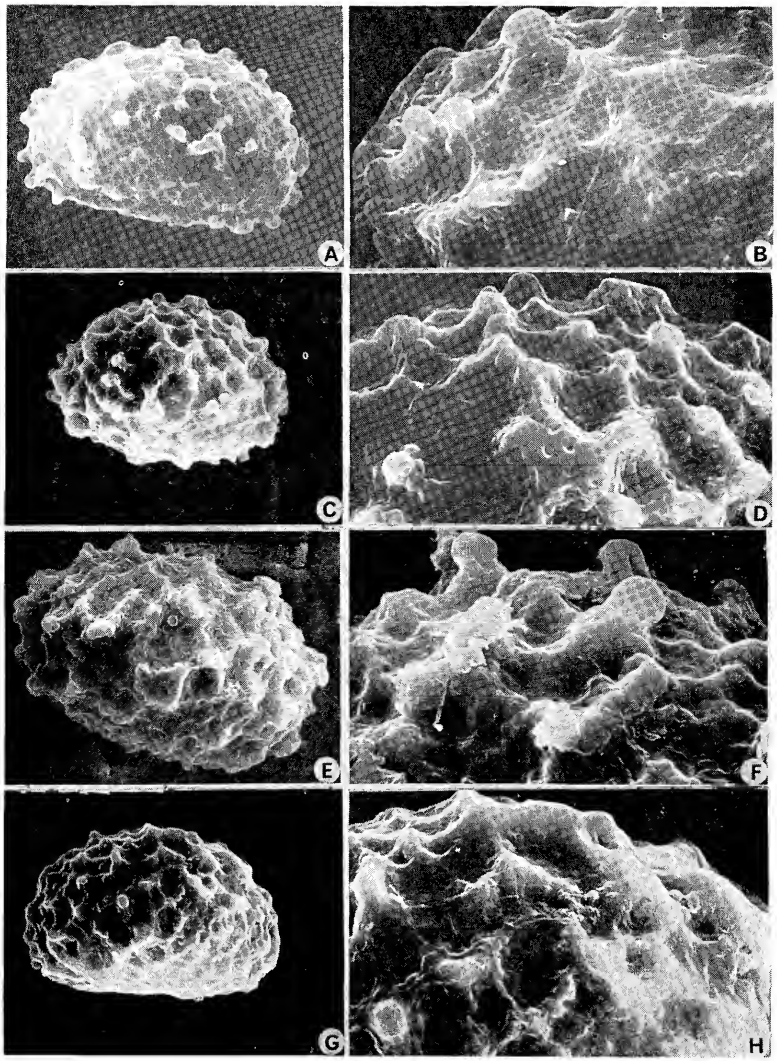
この属は染色体数の面から X=25,26 のグループと X=35 のグループに分けられ，それぞれのグループが形態的な形質で差を示しているが，胞子表面の模様に関しては，この属を二つのグループに分けることは困難なようである。むしろこの属では胞子表面の模様は倍数性と関連がありそうである。



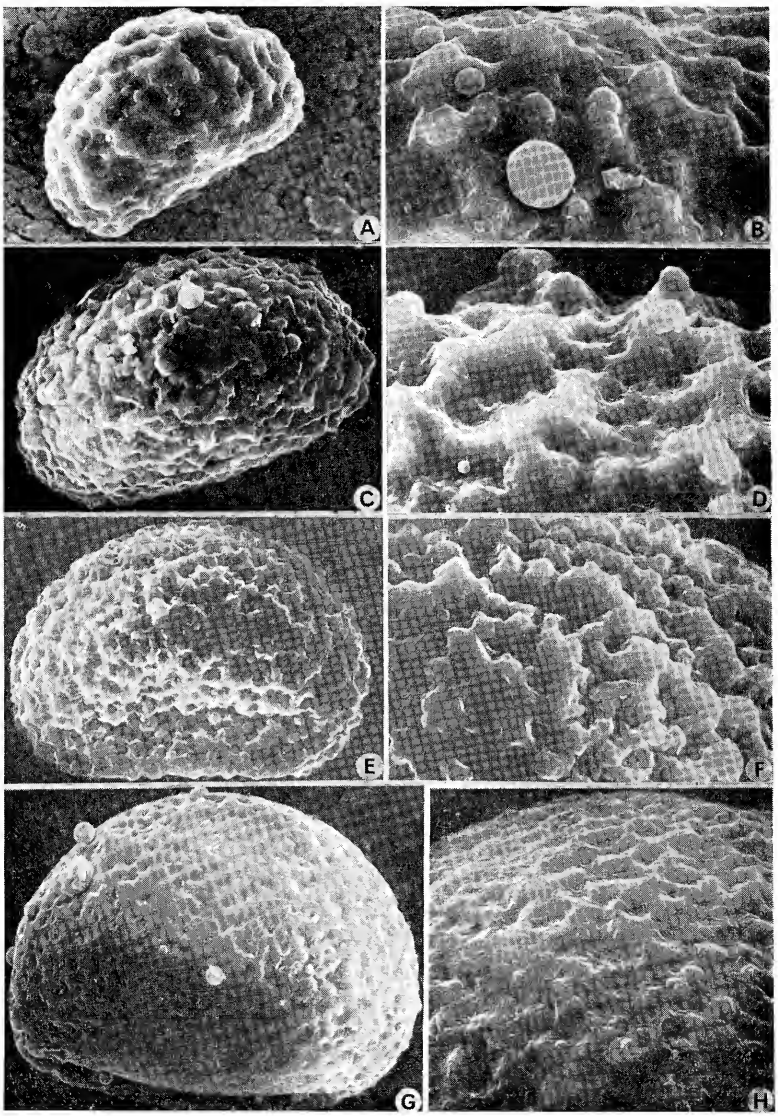
K. MITUI: Spore ornamentations of *Lepisorus*



K. MITUI: Spore ornamentations of *Lepisorus*



K. MITUI: Spore ornamentations of *Lepisorus*



K. MITUI: Spore ornamentations of *Lepisorus*